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Biochar improves soil quality and N_2-fixation and reduces net ecosystem CO_2 exchange in a dryland legume-cereal cropping system

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ABSTRACT

Biochar, a product of the pyrolysis of organic material, has received wide attention as a means to improve soil fertility, crop productivity, and carbon sequestration to mitigate climate change. This study was conducted in an arid zone agricultural region to investigate the impacts of biochar on soil quality, N_2-fixation, and net ecosystem CO_2 exchange (NEE) for a legume-cereal crop rotation (2013-15). Biochar was produced by pyrolysis of sugarcane bagasse and applied at rates of 0%, 0.25%, and 0.5% carbon (C) equivalent, with and without chemical fertilizer (23 kg N, 45 kg P and 25 kg K ha^{-1}). In terms of soil quality, biochar applications increased soil organic carbon (SOC), total nitrogen (TN), and decreased soil bulk density. The yield of mash bean and wheat crop was significantly enhanced in response to biochar treatment with and without chemical fertilizer during both the years of study. N_2-fixation in mash bean increased by 83% in biochar-only (0.5% C) amended soil and by 127% with chemical fertilizer. In the first year of mash bean, no difference was observed in NEE, while a significant decrease was observed in the second year attributed to priming effect of fresh biochar. Wheat NEE decreased by 144% and 200% in the first year, and by 283% and 265% in the second year, in unfertilized soil amended with 0.25 and 0.5% biochar, respectively, as compared to the controls. In treatments with chemical fertilizer, wheat NEE decreased by 311% and 344% in the first year, and by 293% and 292% in the second year, as compared to their respective controls. Biochar treatment increased biomass yield (BY) and grain yield (GY) in both mash bean and wheat crops during both the years with and without chemical fertilizer. These results indicate that sugarcane-bagasse biochar applications in arid soils with low organic carbon may increase crop productivity and N_2-fixation of legumes while reducing the NEE of legume-cereal cropping systems.

1. Introduction

The degree to which biochar soil amendments (BSA) alters soil carbon dioxide (CO_2) emissions likely depends on the priming effect, whereby biochar, or residual labile materials contained in fresh biochar, stimulate the degradation of native soil organic carbon (SOC) (Cross and Sohi, 2011); short-term fluxes of CO_2 can be expected following soil amendment with large amounts of carbon (C) (Steiner et al., 2008; Kuzyakov et al., 2009). Due to its high surface area and porosity, biochar may also stabilize native SOC through direct sorption of dissolved organic matter (Lehmann, 2007). The greatest chemical difference between biochar and other types of organic matter is that biochar consists of a much larger proportion of aromatic C; dominated by innate aromatic C structures (Lehmann et al., 2011). When added to soils, biochar reduces CO_2 emissions due to its recalcitrance and resistance to microbial decomposition (Lehmann, 2007; Shackley et al., 2009; Drake et al., 2016; Zhang et al., 2017). The stability of various biochar products varies, depending on the pyrolysis methodology, temperature, and type of feedstock (Singh et al., 2012). However, the effects of biochar soil amendment on CO_2 flux and soil respiration may vary for different materials and situations (Zimmerman et al., 2011). In a study examining the use of pine wood biochar amendment on a Swiss loamy soil, CO_2 flux was unchanged, whereas CO_2 release increased with the use of a grass-derived biochar (Hilscher et al., 2009). Karhu et al. (2011) reported that birch biochar had no effect on soil CO_2 evolution when applied at the rate of 9 t ha^{-1} to a wheat field in Southern

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Table 1
Soil (0–15 cm) and biochar basic characteristics.

	pH (H_2O)	EC (dS m^{-1})	SOC (g C kg^{-1})	DOC (g C kg^{-1})	TN (g kg^{-1})	Ash content (%)	Bulk density (g cm^{-3})	SMC (%)
Soil	8.25	0.53	5.07	0.46	1.03	/	1.46	9.35
Biochar	6.68	0.30	4.97	4.70	1.40	10.50	/	/

*SOC = Soil organic carbon; DOC = Dissolved organic carbon; TN = Total nitrogen; SMC = Soil moisture content.

Finland, but did increase the soil's water holding capacity up to 11% over the control. Beyond the priming effect, biochar applications have generally been shown to increase stable SOC, which implies its potential to enhance soil C sequestration (Sukartono et al., 2011).

The net ecosystem exchange (NEE) of CO_2 refers to the difference between carbon inputs (carbon assimilation) and carbon outputs (through organic matter decomposition). The NEE values indicate overall CO_2 source activity, related to a shorter period of photosynthetic uptake and metabolic costs of N_2 fixation. Perennial legumes (e.g., alfalfa, *Medicago sativa* L.) are strong sinks for atmospheric CO_2, with an average net ecosystem production of 980 (range 550–1200) (kg ha^{-1}) (Gilmanov et al., 2014). The effects of biochar-amended soil on wheat production have been evaluated in several studies. An oil-Mallee derived biochar, applied at a rate of 6 t ha^{-1} in Pindar, dryland Western Australia, along with half the recommended rate of soluble fertilizer, increased the yield of wheat by 340 kg ha^{-1}, or 18% over the control (Solaiman et al., 2010). Wheat grown with mineral fertilizer and 1.5 t ha^{-1} biochar increased grain yield by 46% over the control. Acacia wood biochar applied at 50 t ha^{-1} increased the productivity and yield of cowpea, mung bean, and *Sesbania* over two cropping seasons in a semi-arid region of Pakistan (Arif et al., 2015). Zhang et al. (2013) reported that biochar application at the rate of 10 t ha^{-1} increased net primary productivity (NPP), while BSA applied at rates of 20 t ha^{-1} and 40 t ha^{-1} exerted large decreases in net greenhouse gas balances (NGHGB) and greenhouse gas intensity (GHGI) of a wheat-rice cropping system.

Globally, biological N_2-fixation (BNF) is currently estimated at 17.2×10^7 tons of N to soils per year, from which leguminous crops added 21.5×10^6 tons approximately half of the total BNF. This demonstrates that BNF is a significant ecosystem service for global agriculture and as such understanding the possible impacts of biochar application on this service is vital. Biochar application to various legumes often enhances biological nitrogen fixation (BNF), with increased nodulation, as shown for white clover, *Trifolium repens* (Rillig et al., 2010), soybean, *Glycine max* (Ogawa and Okimori, 2010), and alfalfa, *Medicago sativa* (George et al., 2012). Possible explanations for enhanced BNF include: (1) low nitrogen (N) availability in soil due to high carbon to nitrogen (C/N) ratios in biochar, resulting in N immobilization (Lehmann et al., 2003); (2) higher pH, EC, organic carbon, and total nitrogen (TN) (Oguntunde et al., 2004); and (3) improved soil physical properties like soil bulk density and moisture content. Various studies (Rondon et al., 2007; Tagoe et al., 2008; Mia et al., 2014) suggested that biochar application might enhance nodulation by increasing BNF with the absorption of compounds involved in N_2-fixation (Nod factors and flavonoids) (Turner, 1955; Thies and Rilling, 2009).

Long-term productivity of the wheat-fallow cropping system depends on the effective use and management of natural resources. Most parts of Pakistan experience a 70- to 80-day hot summer period (air temperatures up to 48 °C) from late April to mid-July, during the cereal-based cropping cycle (between the harvest of wheat and the sowing of maize or other crops). Short duration legume crops can be grown in this "summer gap" as part of a legume-cereal rotation (Arif et al., 2015), which can increase the productivity of a farm by providing additional pulse and oilseed grain legumes, and valued fodder or green manure (Shah et al., 2003; Huang et al., 2017). Retention of nodulated legume residues in soil provides an important source of nitrogen (N) and can increase crop productivity and profitability by promoting greater sustainability of N input into agricultural systems (Peoples et al., 2009; Wu et al., 2016).

Currently, information on CO_2 fluxes and crop productivity in response to biochar C storage for legume-cereal cropping systems is limited. Furthermore, knowledge about N_2 fixation by leguminous crops in biochar-amended soils is scarce. The objectives of this study were to address whether biochar soil amendments affect CO_2 emission over the whole growing period of a legume-cereal cropping system and to evaluate the impacts of biochar in combination with chemical fertilizer on crop yields and N_2-fixation in dryland. A field experiment was conducted to monitor the changes in CO_2 emission after biochar amendment, for mash bean and wheat. Results of this study can assist policymakers in implementing environmentally friendly interventions that employ the use of biochar, as part of an overall strategy to mitigate climate change and ensure food security in arid regions.

2. Materials and methods

2.1. Site description

The study was conducted on a research farm of Pir Mehr Ali Shah-Arid Agriculture University, Rawalpindi, (PMAS-AAUR) Pakistan (33° 1′ N to 36° 6′ N, 73° 30′ E to 73° 45′ E). The soil is a sandy loam with neutral to saline pH (8–8.5), and the area is managed as dryland. The average SOC is less than 1% (Table 1). The climate is semi-arid to subtropical continental, sub-humid and has a bimodal rainfall pattern, with maxima in winter/spring and late summer. Rainfall is erratic, with 60–70% falling during the monsoon wet season (mid-June to mid-September). Meteorological data were taken from Met. Station, Soil and Water Conservation (SAWCRI), Chakwal. Mean monthly temperature, rainfall and evaporation are presented in Fig. 1.

2.2. Biochar production and application

Biochar was produced by pyrolysis of bagasse (sugarcane), using a conventional pyrolysis tank. Sugarcane bagasse biomass was washed to remove the debris and dust particles. The bagasse was air-dried and pyrolysis was performed at 350 °C in an airtight vessel consisting of two metal barrels. The space between the barrels was ignited with natural gas. The process took one hour, and the charcoal was cooled for an additional hour, converting approximately 50% of the biomass to biochar (Gunther, 2009). For the field study, the biochar was ground to pass through a 2 mm sieve and mixed thoroughly to obtain a fine granular consistency that was mixed uniformly with the soil (Pan et al., 2011).

2.3. Field experiment

A two-year (2013–15) field trial was conducted with BSA on a percent carbon basis. Three rates of biochar were amended, with and without chemical fertilizer (NPK), to obtain the following treatments; biochar at 0% C ha^{-1} (B0F0), biochar at 0.25%-C (5 tons) ha^{-1} (B1F0), biochar at 0.5%-C (10 tons) ha^{-1} (B2F0), biochar at 0% C ha^{-1} + NPK (B0F1), biochar at 0.25%-C (5 tons) ha^{-1} + NPK (B1F1), and biochar at 0.5%-C (10 tons) ha^{-1} + NPK (B2F1). The chemical fertilizer was

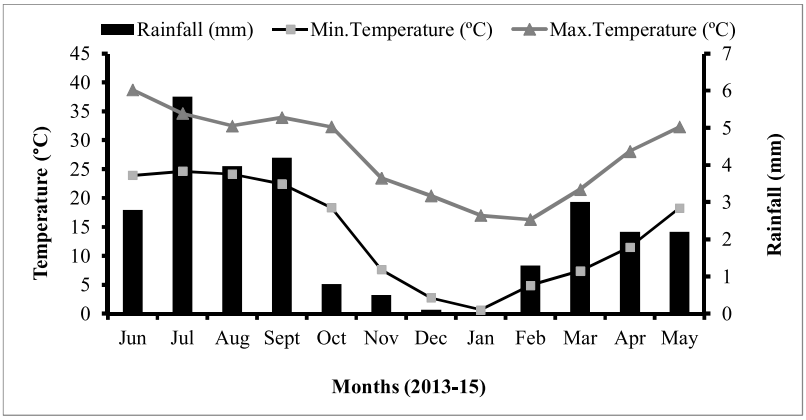

Fig. 1. Average monthly temperature and rainfall during the study period.

applied at the rate of 23 kg N, 45 kg P, and 25 kg K ha^{-1}. Treatments were assigned to field plots (1.5 m × 4.5 m) using a randomized complete block design (RCBD). At the start of experiment, before sowing mash bean, a one-time application of biochar was spread on the surface, thoroughly mixed with soil with a wooden rake, and then plowed to a 15 cm depth. Afterward no biochar was applied to wheat, each treatment was carried out in triplicate plots, and individual plots were separated by border rows of 0.5 m in width. Gas flux was measured at 15 days intervals, whereas crop growth parameters were measured at crop maturity.

2.4. Soil sampling and processing

Composite soil samples were collected at crop maturity at 0–15 cm depth, using a soil core sampler. The samples were preserved in polythene bags and shipped to the laboratory immediately. Root debris was removed; the soil was air-dried and then ground to pass through a 2 mm sieve before analysis.

2.5. Biochar and soil analysis

The biochar and soil moisture contents (SMC) were determined gravimetrically (Gardner et al., 1991). Electrical conductivity (EC) of soil was measured in a saturated paste using a conductivity meter (Rhoades, 1996), and soil pH was determined in soil-distilled water suspensions (ratio of 1:1, w:v) (Thomas, 1996). The pH and EC of the biochar were determined in a mixture of biochar and distilled water (ratio of 1:10, w:v) (Cayuela et al., 2013). Soil and biochar organic carbon, dissolved organic carbon (DOC) and total nitrogen (TN) were determined with a Shimadzu TOC-TN analyzer (Shimadzu Corp., Kyoto, Japan).

2.6. CO_2 flux measurement and net ecosystem CO_2 exchange (NEE)

Evolution of CO_2-C from the soil was measured using a CO_2 analyzer (Lutron GC-2028). For gas flux measurement, a collar was placed in the field plots, and a chamber was placed over the collar at the time of measurement and sealed shut at the joint. Gas flux was measured at 15-days intervals during the growing season. The concentration of CO_2 was analyzed from headspace samples collected at 0, 10, 20 and 30 min (minutes) after closing the chamber. The gas flux was estimated from the slope of the change in CO_2 concentrations CO_2-C (mg $m^{-2} h^{-1}$) in these four samples. The average CO_2 flux and standard deviations (SDs) were calculated from triplicate plots (Zou et al., 2005). The flux of CO_2 was extrapolated to a hectare and converted to kg CO_2-C ha^{-1} for microbial heterotrophic soil respiration (Rh) (Zhang et al., 2010).

The NEE of CO_2 was estimated using the equation given by Zheng et al. (2008):

$$NEE = Rh\text{-}NPP$$

where Rh is the microbial heterotrophic soil respiration (kg-C ha^{-1}) measured by the static chamber method and NPP is the net primary productivity estimated using the equation given by Osaki et al. (1992):

$$NPP = 0.446 \times W_{max} \times -0.00067$$

Where W_{max} is the maximum biomass (Zou et al., 2005).

2.6.1. Crop data

Nodules were counted for five mash bean plants uprooted by excavating a 10 × 10 cm area from each treatment at harvest. Dry weights of the biomass yield (BY) and grain yield (GY) of both crops were determined at harvest. Shoot N (%) was determined with a Shimadzu TOC-TN analyzer (Shimadzu Corp., Kyoto, Japan). N_2-fixation was quantified for mash bean and was estimated by the ^{15}N abundance technique (Europa Scientific 20–20 Stable Isotope Analyzer).

Total N derived from the atmosphere (% N_{dfa}) was estimated using the following equation:

$$\%\, N_{dfa} = \frac{\delta^{15}N(\text{Soil N}) - \delta^{15}N(\text{Legume N})}{(\delta^{15}N(\text{Soil N}) - B)} \times 100$$

where $\delta^{15}N$ (Soil N) is commonly obtained from a non-N_2 fixing reference plant grown in the same soil as the legume, and "B" is the $\delta^{15}N$ of the same N_2 fixing plant when grown with N_2 as the sole source of N.

$$\text{Legume N– uptake (kg ha}^{-1}) = \frac{\text{Legume Total Biomass (kg ha}^{-1}) \times \text{Plant \%N}}{100}$$

$$\text{kg N fixed ha}^{-1} = \text{Legume N uptake (kg ha}^{-1}) \times \%\, N_{dfa} \times 1.5$$

where factor 1.5 is used to include an estimate for the contribution of below-ground N (Shearer and Kohl, 1986; Peoples et al., 1989).

2.7. Statistical analysis

Gas flux and crop growth parameters were analyzed as two-way multi-factorial analysis of variance (ANOVA) using RCBD. The treatment means were compared by using the least significant difference (LSD) test at a significance level $P < 0.05$ (Steel et al., 1997). Abiotic components (pH, EC, BD, SMC, SOC, DOC and TN) were correlated with N_2-fixation using canonical correspondence analysis (CCA).

3. Results

3.1. Impact of biochar on soil properties

Biochar amendments significantly increased SOC, DOC, TN, and SMC. When biochar was applied in the first year to mash bean plots without NPK (B2F0), an increase of 23, 27 and 55% was recorded in

Table 2
Soil bulk density, EC, pH (H_2O), Soil organic carbon (SOC), Dissolved organic carbon (DOC), Total nitrogen (TN) and Soil moisture content (SMC) influenced by biochar application (means ± S.D., n = 3).

2013–14	Treatments	Crop	Bulk density ($g\ cm^{-3}$)	EC ($dS\ m^{-1}$)	pH	SOC ($g\ kg^{-1}$)	DOC ($g\ kg^{-1}$)	TN (%)	SMC (%)
No fertilizer	B0F0	Mash bean	1.46 + 0.02a	0.53 + 0.03a	8.25 + 0.09a	6.07 + 0.42b	0.46 + 0.02f	3.03 + 0.21c	9.35 + 1.42b
		Wheat	1.45 + 0.02a	0.53 + 0.03ab	8.25 + 0.10a	6.20 + 0.20b	0.46 + 0.03c	2.87 + 0.25c	9.17 + 1.81c
	B1F0	Mash bean	1.41 + 0.02bc	0.54 + 0.02a	8.22 + 0.1a	6.59 + 0.05ab	0.48 + 0.02d	3.44 + 0.07bc	12.90 + 1.35ab
		Wheat	1.41 + 0.01ab	0.54 + 0.02ab	8.21 + 0.09a	6.97 + 0.26ab	0.45 + 0.04d	3.06 + 0.04c	11.57 + 1.02ab
	B2F0	Mash bean	1.39 + 0.02c	0.56 + 0.02a	8.11 + 0.27a	7.47 + 0.81a	0.51 + 0.05b	3.86 + 0.36a	14.57 + 1.79a
		Wheat	1.38 + 0.02b	0.56 + 0.01 a	8.11 + 0.27a	8.07 + 0.53a	0.44 + 0.04e	3.65 + 0.40a	12.53 + 0.65ab
NPK Fertilizer	B0F1	Mash bean	1.45 + 0.03ab	0.52 + 0.02a	8.09 + 0.06a	6.20 + 0.60b	0.47 + 0.03e	3.70 + 0.22ab	10.55 + 1.66b
		Wheat	1.42 + 0.05ab	0.52 + 0.01b	8.09 + 0.06a	6.34 + 0.33b	0.48 + 0.02b	3.04 + 0.29c	10.23 + 1.08bc
	B1F1	Mash bean	1.42 + 0.03abc	0.56 + 0.02a	8.18 + 0.15a	6.54 + 1.01ab	0.49 + 0.03cb	3.79 + 0.19ab	11.99 + 3.07b
		Wheat	1.41 + 0.03ab	0.55 + 0.02ab	8.18 + 0.15a	7.80 + 0.61a	0.47 + 0.01a	3.18 + 0.41bc	10.77 + 0.11abc
	B2F1	Mash bean	1.39 + 0.03c	0.54 + 0.01a	8.29 + 0.06a	7.27 + 0.83a	0.52 + 0.04a	3.77 + 0.15ab	12.53 + 1.51ab
		Wheat	1.37 + 0.02b	0.54 + 0.02ab	8.28 + 0.06a	8.17 + 0.59a	0.48 + 0.03	3.55 + 0.36ab	12.77 + 1.76a
2014–15	B0F0	Mash bean	1.42 + 0.03a	0.52 + 0.02ab	8.18 + 0.07a	6.13 + 0.43b	0.46 + 0.02b	3.09 + 0.29c	9.63 + 1.38d
No fertilizer		Wheat	1.41 + 0.02a	0.54 + 0.01 a	8.20 + 0.08a	6.20 + 0.57c	0.47 + 0.04b	3.19 + 0.20ab	8.72 + 0.65e
	B1F0	Mash bean	1.40 + 0.02ab	0.53 + 0.01 ab	8.20 + 0.05a	6.83 + 0.29ab	0.45 + 0.04c	3.38 + 0.26bc	12.44 + 0.98bc
		Wheat	1.39 + 0.02ab	0.53 + 0.01a	8.22 + 0.04a	7.12 + 0.53b	0.45 + 0.03c	2.93 + 0.23c	12.77 + 1.19c
	B2F0	Mash bean	1.37 + 0.01b	0.54 + 0.01 a	8.23 + 0.06a	7.57 + 0.85a	0.44 + 0.03d	3.56 + 0.27ab	13.59 + 0.41ab
		Wheat	1.35 + 0.02 cd	0.55 + 0..02a	8.14 + 0.05a	8.13 + 0.49a	0.44 + 0.03d	3.17 + 0.13abc	14.17 + 0.60ab
NPK Fertilizer	B0F1	Mash bean	1.43 + 0.02a	0.51 + 0.01 b	8.12 + 0.13a	6.29 + 0.62b	0.48 + 0.03a	3.25 + 0.30bc	11.25 + 1.03 cd
		Wheat	1.42 + 0.02a	0.52 + 0.01a	8.10 + 0.11a	6.30 + 0.54c	0.48 + 0.05a	3.30 + 0.20a	11.32 + 0.96d
	B1F1	Mash bean	1.40 + 0.03ab	0.52 + 0.02 ab	8.14 + 0.12a	6.67 + 1.08ab	0.46 + 0.04b	3.57 + 0.27ab	12.31 + 1.41bc
		Wheat	1.38 + 0.03bc	0.52 + 0.01a	8.12 + 0.11a	7.21 + 0.48b	0.47 + 0.04b	2.98 + 014bc	13.51 + 1.05bc
	B2F1	Mash bean	1.36 + 0.02b	0.53 + 0.02 ab	8.22 + 0.03a	7.48 + 0.70a	0.48 + 0.03a	3.87 + 0.43a	14.51 + 1.06a
		Wheat	1.36 + 0.02d	0.53 + 0.03a	8.16 + 0.06a	8.29 + 0.46a	0.48 + 0.05a	3.20 + 0.16ab	15.02 + 1.28a

Biochar amendment at 0% C, 0.25% C and 0.5% C ha^{-1} (B0, B1 and B2, respectively) with (F1) and without NPK fertilization (F0).
Letters in a single column indicate a statistical difference among the treatments at $P < 0.05$. Means with the same small letters are not significantly different at $P < 0.05$. It represents biochar × NPK fertilization interaction effects.

SOC, DOC, and TN, respectively, as compared to the control (B0F0) plots (Table 2). However, soil bulk density was reduced after biochar application. When biochar was added to mash bean plots along with NPK, SOC and SMC increased by 17 and 19%, respectively. The same trend observed in the first year was recorded in mash bean plots during the second year, except for a slight overall decrease in soil BD and EC. The wheat crop in the B2F0 treatment plots in its first year showed an increase of 30, 27 and 37% in SOC, TN, and SMC, respectively, except for DOC, which decreased significantly. A similar trend in these soil parameters was also recorded during the second year for the wheat crop.

3.2. CO_2 flux as influenced by biochar

In the mash bean field experiment, CO_2 emissions declined significantly over time (days), with 47 (73 mg CO_2-C $m^{-2}\,h^{-1}$) and 34% (91 mg CO_2-C $m^{-2}\,h^{-1}$) decline was observed on Oct 24 and Oct 10, respectively, in contrast to 04-Jul (137 mg CO_2-C $m^{-2}\,h^{-1}$) (2013–14) (Table 3). The interactive effect of biochar and days showed a maximum CO_2 flux with biochar application at first day of application (04-July) (i.e., 143 mg CO_2-C $m^{-2}\,h^{-1}$), while the lowest CO_2 flux was observed on 24-Oct without biochar (i.e., 65 mg CO_2-C $m^{-2}\,h^{-1}$), indicating a 55% decrease in comparison to 04-Jul. Emissions of CO_2 in the B2 level of biochar differed significantly from levels B1 and B0, and also differed significantly in both levels of fertilizer (F0 & F1); however, the biochar × fertilizer × day's interaction revealed non-significant differences in CO_2 emissions (Fig. 2).

Emissions of CO_2 also differed significantly with time (days) in the wheat experiments, and showed a 67% decline on 21-Jan (29 mg CO_2-C $m^{-2}\,h^{-1}$), and a 45% on 04-Feb (49 mg CO_2-C $m^{-2}\,h^{-1}$), in contrast to emissions on 30-Apr (89 mg CO_2-C $m^{-2}\,h^{-1}$) (Table 3). The fertilizer × days interaction exhibited the highest CO_2 flux without NPK on 30-Apr (i.e., 89 mg CO_2-C $m^{-2}\,h^{-1}$), while the lowest CO_2 flux was observed on 21-Jan with NPK (i.e., 25 mg CO_2-C $m^{-2}\,h^{-1}$), indicating a 72% drop in emissions. However, the levels of CO_2 emissions differed significantly between both levels of fertilizer (F0 and F1) and all levels of biochar tested (B0, B1, and B2). In treatments without chemical fertilizer application, the biochar × fertilizer × days interaction revealed significant differences in emissions observed in B2F0 and B1F0, when compared to those in B0F0 (Fig. 2). The maximum CO_2 emission (106 mg CO_2-C $m^{-2}\,h^{-1}$) was observed in B0F0 on 30-Apr, followed by 15-Apr (96 mg CO_2-C $m^{-2}\,h^{-1}$), in comparison to that observed in B1F0 on 07-Jan (i.e. 20 mg CO_2-C $m^{-2}\,h^{-1}$). Likewise, with NPK, emissions in B2F1 and B1F1 differed significantly from those observed in B0F1; however, the biochar application rate exhibited non-significant effects on soil respiration. The same trend in CO_2 emissions was noted for both crops in 2014-15, except that the emissions were reduced in both crops during the second growing season (Table 4) (Fig. 3). No priming effect of biochar was noticed in the second year, while fertilizer application yielded higher carbon loss in terms of carbon emissions.

3.3. Net ecosystem CO_2 exchange and crop yield

Results of heterotrophic soil respiration (Rh) (kg-C ha^{-1}), NEE (kg-C ha^{-1}) and crop yield (kg ha^{-1}), are presented in Table 5. Biochar applications to soil significantly decreased Rh in wheat, regardless of NPK fertilization; however, no significant variation in Rh was observed in mash bean in 2013. In the first year of the wheat field experiments, biochar application without NPK decreased Rh by 31 and 17% in unfertilized soil amended with 0.25% and 0.5% biochar, respectively, and by 27 and 26% in plots treated with chemical fertilizer, as compared to their respective controls. Similar results were obtained for wheat during the second year, but with greater declines in Rh values (43 and 46% declines), with and without chemical fertilizer, respectively as compared to the controls (B0F0 and B0F1). The NEE for wheat decreased by 147 and 200% in the first year, and 283 and 265% in the second year, in unfertilized soil amended with 0.25 and 0.5% biochar, respectively. In treatments with chemical fertilizer, the NEE for wheat decreased by 311 and 344% in the first year and 293 and 292% in the second year, as compared to their respective controls. No significant difference was observed for NEE in the first year of the mash bean crop; however, a significant decrease in NEE was observed in the second season.

Table 3
Treatment (Biochar) and factor (Fertilizer) effects on carbon dioxide (g CO_2 m^{-2} h^{-1}) flux on mash bean-wheat (2013-14).

Mash bean	Days (2013-14)									
Fertilizer	04-Jul	18-Jul	01-Aug	15-Aug	29-Aug	12-Sep	26-Sep	10-Oct	24-Oct	Mean
F0	136ab	131cd	125ef	118gh	111i	101jk	94lm	88n	68p	108b
F1	139a	133bc	127de	121fg	115hi	105j	98kl	94m	77o	112a
Biochar										
B0	133bc	127cde	123ef	116gh	110ij	105jk	101k	93mn	83o	110b
B1	137b	132bc	125def	121fg	113hi	100kl	92mn	87no	65q	108b
B2	143a	136b	129cd	121fg	115hi	104k	95lm	91mn	71p	112a
Mean	137	132	125	119	113	103	96	91	73i	

Wheat	Days (2013-14)													
Fertilizer	29-Oct	12-Nov	26-Nov	10-Dec	24-Dec	07-Jan	21-Jan	04-Feb	18-Feb	04-Mar	18-Mar	01-Apr	15-Ap	30-Ap	Mean
F0	82b	75c	66ef	64f	54gh	28ij	25j	49h	67def	49h	72cd	71cd	83b	87ab	62 b
F1	87ab	76 c	72cd	65ef	52gh	32 i	33i	50h	63f	56g	70cde	75c	84ab	89 a	65 a
Biochar															
B0	89cd	84d-g	81fgh	66k-n	58o-r	38st	39s	79gh	56pqr	86c-f	88cde	91c	101b	108a	76a
B1	80fgh	69jkl	61n-q	63m-p	45s	22x	24v-x	29u-w	64l-o	38st	53r	58o-r	72ijk	75hij	54c
B2	84d-g	72ijk	63l-p	64l-o	55qr	30uv	23wx	39s	75hij	32tu	72ijk	69j-m	77g-i	81e-h	59b
Mean	84b	75c	68ef	64g	53h	30i	29i	49h	65fg	52h	71de	73cd	83b	89a	

Biochar amendment at 0% C, 0.25 %C and 0.5 %C ha^{-1} (B0, B1 and B2) respectively, with (F1) and without NPK fertilization (F0).

Biochar application to soil significantly enhanced the BY and GY of both crops. During the first year of the mash bean study, treatment plots without NPK (B2F0) showed an increase of 24, and 7% in BY and GY, respectively, as compared to the control (B0F0) plots. When biochar was added along with NPK to mash bean plots, BY and GY increased by 6 and 9%, respectively. The same trend that was observed during the first year of the mash bean study, was recorded during its second year, except for an overall slight decrease in BY and GY during the second year. During the first year of the wheat crop, an increase of 6 and 12% was observed in BY and GY, respectively, in the treatment without fertilizer (B2F0) and compared to B0F1. In plots that co-used biochar and NPK fertilizer, an increase of 23 and 11% was recorded in BY and GY, respectively, as compared to B0F1. A similar trend in BY and GY was observed during the second year of the wheat study.

3.4. Biochar influence on nodule number (NdN) and N_2-fixation

Biochar applications in mash bean plots over both growing seasons significantly enhanced nodule number, shoot N content, N uptake, and N_2-fixation, with and without NPK (Table 6). In treatments without

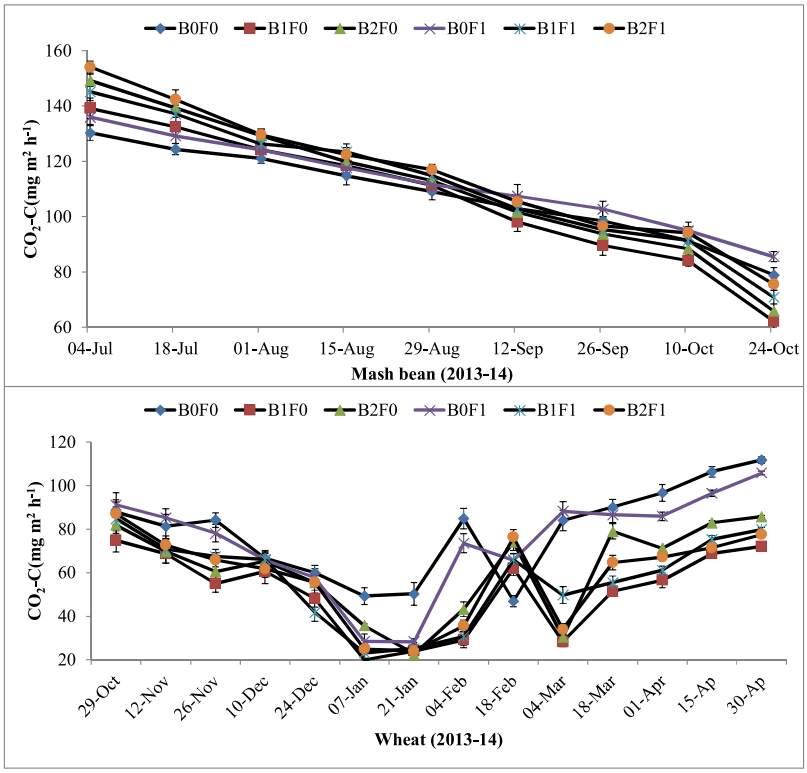

Fig. 2. Effect of biochar and fertilization over time (days) on CO_2 flux during the first growing season (2013-14). Biochar amendment at 0% C, 0.25% C and 0.5% C ha^{-1} (B0, B1 and B2, respectively) with (F1) and without NPK fertilization (F0).

Table 4
Treatment (Biochar) and factor (Fertilizer) effects on carbon dioxide (g CO_2 m^{-2} h^{-1}) flux on mash bean-wheat (2014-15).

Mash bean	Days (2014-2015)									
Fertilizer	04-Jul	18-Jul	01-Aug	15-Aug	29-Aug	12-Sep	26-Sep	10-Oct	24-Oct	Mean
F0	107abc	105bcd	98de	101cde	104 bcd	94ef	87fg	78h	69i	94b
F1	114a	111ab	110 ab	105bcd	102cd	94ef	85gh	83gh	83gh	99a
Biochar										
B0	123a	115abc	107c-f	113 bcd	119ab	112bcd	104d-h	96hij	89jk	108a
B1	99f-i	98f-i	98ghi	94 hij	9ij	82kl	75lmn	74lmn	66n	86b
B2	110b-e	110b-e	106c-g	101e-h	98ghi	88jk	79lm	79lm	72mn	94c
Mean	111a	108 ab	104 b	10 b	103 b	93 c	85 d	83 d	75 e	

Wheat	Days (2014-2015)													
Fertilizer	29-Oct	12-Nov	26-Nov	10-Dec	24-Dec	07-Jan	21-Jan	04-Feb	18-Feb	04-Mar	18-Mar	01-Apr	15-Ap	30-Ap	Mean
F0	82b	67d	54ef	54ef	45hi	25l	33k	35jk	53ef	45hi	53efg	57e	72cd	78bc	**54b**
F1	80b	68d	65d	65d	50fgh	36jk	46ghi	41ij	45hi	54ef	67d	66d	78bc	89a	**61a**
Biochar															
B0	91cd	77efg	77fg	70ghi	66hi	44q-t	54m-p	75fgh	64ij	85cde	91cd	92c	116b	125a	**81a**
B1	69ghi	62i-m	48o-r	56j-n	35u-y	21a	28x-a	24za	40r-v	32w-z	42r-u	48n-r	55k-o	63i-k	**44c**
B2	83def	62i-l	53nop	53n-q	40r-v	27yza	36t-x	38s-w	43 r-u	32v-z	45p-s	44q-t	54l-p	63 i-l	**48b**
Mean	**81a**	**67c**	**59d**	**60d**	**47e**	**31g**	**40f**	**38f**	**49e**	**50e**	**60d**	**62d**	**75b**	**84a**	

Biochar amendment at 0% C, 0.25% C and 0.5%C ha^{-1} (B0, B1 and B2) respectively, with (F1) and without NPK fertilization (F0).

NPK and in comparison to treatments without biochar over the first season, the nodule number, shoot N content, N uptake and N_2-fixation increased by 89, 26, 51 and 83%, respectively with an application rate of 0.5% C (B2F0), and by 27, 8, 247 and 83%, respectively with biochar applied at 0.25% C (B1F0). In contrast, NPK fertilization improved shoot nodule number, N content, total N uptake and N_2-fixation up to 21, 95, 66 and 127% respectively under biochar application at 0.5% C (B2F0), and by 13, 52, 4 and 120% respectively with biochar at 0.25% C (B1F0), as compared to NPK alone (B0F1). Lower values of nodule number, shoot N content, N uptake and N_2-fixation were observed during the second season

3.5. Canonical correspondence analysis (CCA)

CCA was performed on soil variables and N_2-fixation indices (Fig. 5). The total variance explained by the first two axes in 2013 was

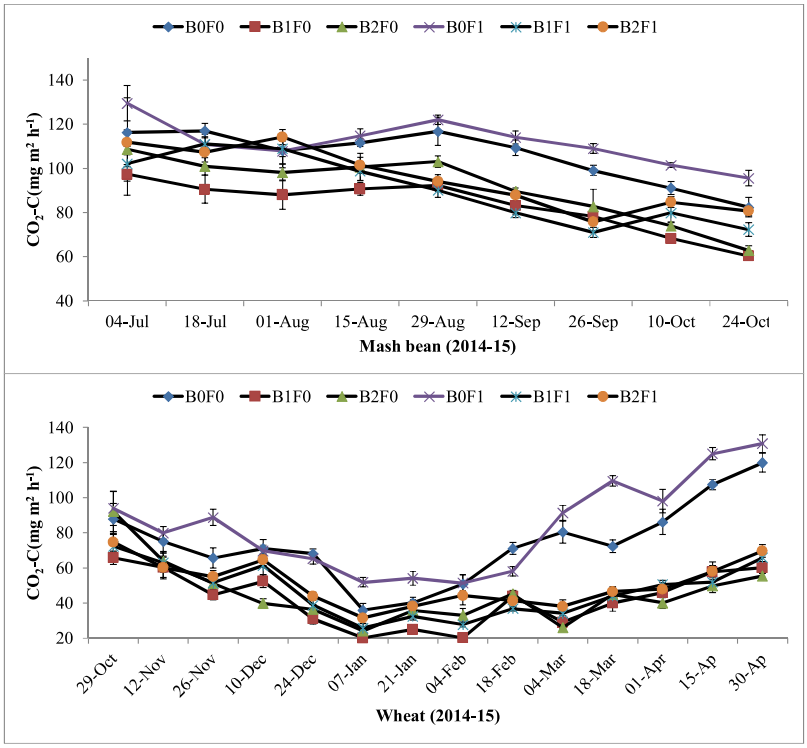

Fig. 3. Effect of biochar and fertilization over time (days) on CO_2 flux during the second growing season (2014-15). Biochar amendment at 0% C, 0.25% C and 0.5% C ha^{-1} (B0, B1 and B2, respectively) with (F1) and without NPK fertilization (F0).

Table 5
The net ecosystem CO_2 exchange (NEE) and crop yield of the legume-cereal cropping system.

2013–14	Crop	Treatments	Rh (kg-C ha^{-1})	NPP (kg-C ha^{-1})	NEE (kg-C ha^{-1})	BY (kg ha^{-1})	GY (kg ha^{-1})
No fertilizer	Mash Bean	B0F0	2930 ± 32 cd	1219 ± 76e	1820 ± 78a	2649 ± 17.1e	612 ± 3.7d
	Wheat		3255 ± 98b	2763 ± 134d	492 ± 201a	6124 ± 29.4e	2178 ± 7.5e
	Mash Bean	B1F0	2876 ± 98d	1394 ± 75d	1658 ± 32b	3030 ± 15.3d	636 ± 0.66 cd
	Wheat		2252 ± 141e	2745 ± 86d	−493 ± 70c	6374 ± 23.9d	2312 ± 16.2de
	Mash Bean	B2F0	2981 ± 21bc	1505 ± 72c	1638 ± 56b	3272 ± 14.4 cd	656 ± 3.9 cd
	Wheat		2690 ± 24c	2921 ± 163d	−231 ± 156b	6488 ± 28.3d	2443 ± 8.3 cd
NPK Fertilizer	Mash Bean	B0F1	3041 ± 12ab	1571 ± 59bc	1565 ± 78b	3415 ± 16.0bc	690 ± 2.0bc
	Wheat		3455 ± 77a	3045 ± 137c	411 ± 211a	6826 ± 25.8c	2617 ± 7.2bc
	Mash Bean	B1F1	2997 ± 55abc	1604 ± 79ab	1554 ± 110b	3485 ± 15.7ab	721 ± 3.0ab
	Wheat		2498 ± 25d	3366 ± 184b	−867 ± 203d	7984 ± 29.2b	2764 ± 8.5ab
	Mash Bean	B2F1	3079 ± 34a	1660 ± 66a	1560 ± 106b	3609 ± 13.5a	751 ± 2.7a
	Wheat		2567 ± 27 cd	3570 ± 179a	−1002 ± 154d	8375 ± 28.4a	2894 ± 18.8a
2014–15	Mash Bean	B0F0	2864 ± 54b	1207 ± 79e	1657 ± 36a	2625 ± 171 e	622 ± 19 e
No fertilizer	Wheat		3273 ± 101b	2731 ± 131e	541 ± 188b	6195 ± 300d	2062 ± 257c
	Mash Bean	B1F0	2257 ± 112e	1374 ± 70d	882 ± 122b	2987 ± 153d	659 ± 24de
	Wheat		1855 ± 16e	2843 ± 107de	−988 ± 121c	6154 ± 192d	2202 ± 234c
	Mash Bean	B2F0	2483 ± 19d	1454 ± 66 cd	1029 ± 72b	3160 ± 144 cd	681 ± 30 cd
	Wheat		2002 ± 79d	2894 ± 126d	−892 ± 204c	6549 ± 366c	2519 ± 257 b
NPK Fertilizer	Mash Bean	B0F1	3054 ± 26a	1521 ± 74bc	1534 ± 91a	3305 ± 160bc	715 ± 23bc
	Wheat		3822 ± 144a	3029 ± 147c	777 ± 239a	6826 ± 306c	2716 ± 271 b
	Mash Bean	B1F1	2441 ± 70d	1572 ± 72ab	869 ± 141b	3417 ± 157ab	750 ± 30ab
	Wheat		2058 ± 52d	3561 ± 131b	−1503 ± 89d	7547 ± 412b	2561 ± 237b
	Mash Bean	B2F1	2612 ± 14c	1624 ± 62b	988 ± 53b	3531 ± 135 a	782 ± 19a
	Wheat		2240 ± 44c	3735 ± 127a	−1495 ± 86d	8003 ± 402a	2954 ± 357 a

Biochar amendment at 0% C, 0.25% C and 0.5% C ha^{-1} (B0, B1 and B2, respectively) with (F1) and without NPK fertilization (F0).
Letters in a single column indicate a statistical difference among the treatment means at $p < 0.05$. Means with the same small letters are not significantly different at $p < 0.05$. It represents biochar × NPK fertilization interaction effects. *Rh = Heterotrophic soil respiration; NPP = Net primary productivity; NEE = Net ecosystem exchange; BY = Biological yield; GY = Grain yield.

Table 6
The effect of biochar application on nodule number in mash bean and N_2 fixation.

	Treatment	Season	Nodule number	% Shoot N	% $^{a}N_{dfa}$	N uptake (kg ha^{-1})	N_2-Fixed (kg ha^{-1})
No fertilizer	B0F0	2013	37 ± 6d	1.43 ± 0.04c	104 ± 5bc	38 ± 1d	58 ± 5d
		2014	33 ± 4c	1.58 ± 0.08b	95 ± 7b	42 ± 18c	60 ± 3d
	B1F0	2013	47 ± 8c	1.55 ± 0.05bc	117 ± 3abc	46 ± 2 cd	81 ± 4c
		2014	43 ± 8c	1.67 ± 0.13ab	95 ± 50b	51 ± 4bc	72 ± 9 cd
	B2F0	2013	70 ± 7b	1.80 ± 0.33b	127 ± 9ab	57 ± 8b	107 ± 8b
		2014	61 ± 11b	1.81 ± 0.26ab	96 ± 5b	59 ± 11b	86 ± 20bc
NPK Fertilizer	B0F1	2013	68 ± 3b	1.16 ± 0.04c	98 ± 26c	48 ± 2bc	70 ± 16 cd
		2014	60 ± 5b	1.64 ± 0.12ab	77 ± 8c	56 ± 6b	65 ± 12 cd
	B1F1	2013	77 ± 2ab	2.18 ± 0.19a	140 ± 17a	74 ± 5.7a	155 ± 10a
		2014	70 ± 2ab	1.73 ± 0.17ab	118 ± 8a	60 ± 3ab	107 ± 11ab
	B2F1	2013	82 ± 7a	2.26 ± 0.08a	133 ± 10a	80 ± 6a	159 ± 23a
		2014	76 ± 7a	1.95 ± 0.15a	113 ± 7a	70 ± 8a	119 ± 15a

Biochar amendment at 0, 0.25 %C and 0.5 %C ha^{-1} (B0, B1 and B2, respectively) with (F1) and without NPK fertilization (F0).
Letters in a single column indicate a statistical difference among the treatments at $p < 0.05$. Means with the same small letters are not significantly different at $p < 0.05$. It represents biochar × NPK fertilization interaction effects.
[a] N_{dfa} = total N derived from the atmosphere.

70.59% (axis-1) and 28.4% (axis-2). Indices of TN (0.9), DOC (0.81), SOC (0.61), and EC (0.52) were best correlated with axis-1, and pH (0.72) with axis-2. The N_2-fixation indices are regarded to be significant when their values exceed 1; thus, significant variables included the amount of N_2 fixed (N-fixed in Fig. 5), and shoot N (its dominant driver was a combination of DOC, TN, SOC, and EC). In 2014, significant variables were N_{dfa} and N_2-fixed (its dominant driver was a combination of TN, SMC, and SOC).

4. Discussion

4.1. Effect of biochar on soil properties

Physical and biochemical properties of soil have been shown previously to change with biochar amendment (Asai et al., 2009; Major et al., 2010). Different crops respond differently to the physicochemical properties of biochar, in addition to soil type and climatic conditions (Haefele et al., 2011). Biochar soil application increased the porosity and water holding capacity of the soil and decreased its bulk density (Ogawa and Okimori, 2010), which promoted lateral root formation and an increase in soil volume that was exploited by plant roots. The results of our study revealed that biochar application improved the soil's physical and chemical properties (Table 2); these results are supported by the studies of Asai et al. (2009) and Major et al. (2010). Chen et al. (2010) reported that bagasse charcoal reduced soil dry density and increased the moisture content of Shimajiri Maji soil. In contrast, biochar application increased the SOC and TN, since it contained 49% C. The higher TN is likely due to reduced NO_3 leaching, and NH_4 adsorption to biochar particles (Paz-Ferreiro et al., 2017). These results are supported by Blackwell et al. (2010), who reported improved soil physicochemical properties and better crop growth with a biochar application of 10 t ha^{-1}. The enhanced soil moisture content (SMC) observed in this study is likely due to the large surface area of biochar particles, and increased pore availability, both of which promote the

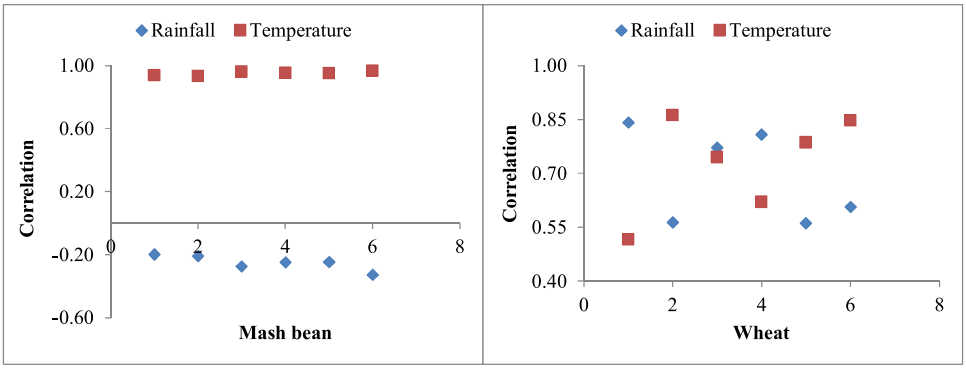

Fig. 4. Correlation of CO_2 flux (CO_2-C ha^{-1} $h^{-1)}$ with rainfall (mm) and temperature (°C) in the mash bean-wheat growing cycle.

water holding capacity of biochar particles. In this study, biochar increased soil water content and reduced its bulk density in the second year after application; these results are also consistent with Tammeorg et al. (2014). A meta-analysis study (Biederman and Harpole, 2013) also confirms our results, indicating that biochar applications to soils enhance crop productivity and limits nutrient leaching. Bagasse biochar benefits soils by reducing fertilizer requirement and improving crop growth. These effects are likely attributed to enhanced essential nutrient and water uptake promoted by biochar in less fertile soils of the arid region.

4.2. N_2-fixation and crop yield

Biochar has been proven to improve soil physicochemical properties and to increase the abundance of free-living and symbiotic microorganisms involved in N_2-fixation. In our study, biochar enhanced nodule number and improved BNF with and without chemical fertilizer application. Many soil quality factors can affect the degree of legume nodulation, including soil bulk density, pH, carbon, and nitrogen availability (Slattery et al., 2001), all of which influenced by biochar application (Jones et al., 2012). The improved BNF observed is most likely due to a combination of factors related to enhanced nodule number, nutrient availability in soil (Lehmann et al., 2003), and stimulation of plant-microbe interactions (Saito and Marumoto, 2002). CCA revealed that pH, EC, SOC, DOC, TN, and BD are best correlated with N_2-fixation (Fig. 5). Availability of carbon in the form of DOC at the start of the experiment resulted in improved nitrogen fixation, by likely enhancing the growth of nitrogen-fixing organisms present in the soil. Decreased BD also enhances soil pore space, which in turn promotes root growth. Increased pore space and nutrient availability also improve root nodulation (Paredes et al., 2008). Nishio and Okano (1991) reported that BNF was 15% higher when biochar was added to soil at the early stages of alfalfa development, and 227% higher when applied at peak nodule development.

Possible explanations for enhanced BNF include: (1) low N availability in soil due to high C/N ratios in biochar, resulting in N immobilization (Lehmann et al., 2003); (2) SOC, DOC, and TN (Oguntunde et al., 2004); and (3) improved soil physical properties like BD and SMC. Mia et al. (2014) reported that biochar applications up to 10 t ha^{-1} led to enhanced BNF, nodule number, and total biomass. Quilliam et al. (2013) also reported enhanced BNF in the presence of biochar. Biochar enhances BNF by stimulating nodulation via absorption of different compounds such as like flavonoids and Nod factors (Thies and Rilling, 2009; Turner, 1955). Rondon et al. (2007) and Tagoe et al. (2008) found similar results in common beans and in soybean grown in soils treated with biochar at higher rates (78 t ha^{-1} and 100 t ha^{-1}). In comparison to the first year, dissolved organic carbon remained unchanged during the second year. Secondly, the total nitrogen and nodule numbers were also lower during the second growing cycle. The positive priming effect may be the possible explanation for the higher nitrogen fixation as labile carbon portion of the biochar which may increase the microbial community involved in the BNF. Differences observed in the results of previous studies of biochar applications are likely due to differences in biochar properties, soil nutrient status, and/or varying responses of plant species to biochar application.

Plant growth is also affected by biochar-induced changes in soil nutrient cycles, particularly the cycling of carbon and nitrogen (Taghizadeh-Toosi et al., 2012). Biochar improves the water holding capacity of the soil, which enhances the biomass and grain yield of both crops (Laird et al., 2010; Kammann et al., 2011). Improved physicochemical properties of soil, coupled with N_2-fixation, resulted in enhanced crop yields, especially in the mash bean crop. Vaccari et al. (2011) reported similar findings in coppice woodlands, where wheat yields were enhanced by 28% and 29% in a silty loam soil in the first growing cycle, and by 32% and 24% in the second cycle. Alburquerque et al. (2013) also reported a 20–30% increase in wheat yield with applications of biochar (made from wheat straw and olive tree prunings) on the loamy sand, which enhanced nutrient availability in the soil.

4.3. CO_2 emission and NEE

Our study shows that biochar applications can stimulate CO_2 emission, likely as a result of positive priming at the start of the experiment (Fig. 2). Lower temperature and less rainfall in the winter season caused lower CO_2 emissions in the wheat crop, while a higher CO_2 flux was correlated with high temperature (Fig. 4). High temperature and moisture availability boost the microbial activity by utilizing the soil organic carbon via organic carbon mineralization (Wang et al., 2013; Hopkins et al., 2014). Moreover, we found that the observed positive priming of biochar is related to its lability (DOC) (Table 2). Biochar application increased labile carbon in soils with mash bean, while reducing DOC content in soils with wheat. Some studies have reported that even trace amounts of low molecular weight organic substrates (e.g. glucose, amino acids, and root exudates) stimulate microbial biomass and activity, and consequently induce 'apparent' or 'real' positive priming effects (Singh and Cowie, 2014). Biochar may contain small quantities of water-soluble low molecular weight organic compounds dispersed among predominantly complex C substrates, which could trigger priming effects (Singh et al., 2012). Our positive priming results are consistent with the findings of Luo et al. (2013), who reported greater positive priming effects with low temperature (350 °C) biochar. Similarly, Farrell et al. (2013) recorded a short-period (up to 9 days) pulse of CO_2 emissions from positively primed soil in the presence of biochar, followed by a decrease in the magnitude of positive priming that persisted throughout the 74 days of incubation. In comparison with the chemical fertilizer, higher CO_2 emission was observed during both years. Hydrolysis of the urea is also involved in enhancing CO_2 emissions. Fertilizer application accelerates decomposition of the native SOC by improved crop growth and

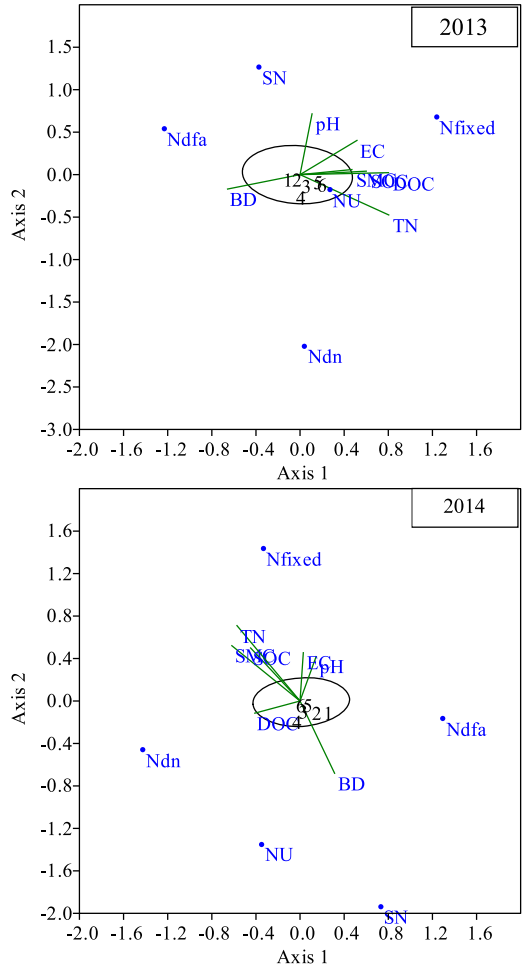

Fig. 5. Canonical correspondence analysis of abiotic component on nitrogen fixation indices.

**EC = Electrical conductivity; SMC = Soil moisture content; BD = Bulk density; SOC = Soil organic carbon; DOC = Dissolved organic carbon; TN = Total nitrogen

***Ndn = Nodule number; NU = Nitrogen uptake; Ndfa = total N derived from the atmosphere

improved substrate (e.g. crop-derived C) and nutrient supply for microbes, thus stimulating microbial activity and enhancing SOC decomposition (Gong et al., 2012).

Importantly, in terms of long-term measurements (in the second growing season), our study recorded a significant decrease in soil CO_2 flux across all biochar treatments (Fig. 3). This reduction in CO_2 flux at later stages may have resulted from, (1) depletion of labile C due to previous positive priming (SOC depletion hypothesis) (Lehmann et al., 2011) and/or (2) stabilization of SOC due to biochar-induced organo-mineral interactions (SOC stabilization hypothesis) (Spokas et al., 2011) which become prominent as labile biochar components are depleted, and as biochar ages over time. Stabilization of native SOC via biochar-induced organo-mineral interactions has been hypothesized to occur in anthrosols containing aged biochar and kaolinite-dominated clay minerals, where the newly added C in sugarcane residues was rapidly stabilized in organo-mineral fractions (Guenet et al., 2010). Using the same biochar as that in the present study, Keith et al. (2011) found that sugarcane residue C applied in the presence of biochar was stabilized rapidly, and this negative priming effect increased with increasing input of residue-C. Similarly, the high surface area and porosity of biochar may also stabilize native SOC through direct sorption of DOC (Lehmann et al., 2011). The sugarcane-bagasse biochar contained more nitrogenous functional groups (Creamer et al., 2014), which likely promotes the physisorption of CO_2 on biochar surfaces due to the strong interaction between the acidic CO_2 and basic nitrogenous surface functional groups (Chang et al., 2013).

The net ecosystem exchange of CO_2 refers to the balance between C outputs from soil heterotrophic respiration (through organic matter decomposition), and C inputs by autotrophic C assimilation (through plant photosynthesis). NEE was determined using the static chamber method, similar to previous studies (Nishimura et al., 2008 and Zhang et al., 2013). In mash bean, the Rh and NPP were higher due to number of factors, 1) priming effect due to fresh biochar application; 2) high temperature; 3) high rainfall during mash bean growing cycles leads to increased soil respiration and 4) improved soil physical properties resulted in enhanced crop productivity which leads no differences in NEE. The levels of NEE, Rh and NPP assessed in wheat are lower than those reported by Liu et al. (2012a; 2012b) and Zhang et al. (2013). In our study, the NEE of control plots was 2.4 t C ha^{-1} higher than that observed in the whole mash-wheat cropping pattern. Zhang et al. (2013) reported that NEE from a wheat-rice rotation cycle was -8.9 t C ha^{-1}, which was greater than the quantity measured in our study, likely due to the different cropping systems. The low NEE observed in our study, in contrast to previous studies, was due to the low CO_2 flux and net primary productivity in our plots. Gilmanov et al. (2014) reported that annual legume crops demonstrated a broad range of net ecosystem production, from 760 to −2100 (kg ha^{-1}), with an average of –330 (kg ha^{-1}). These NEE values indicate an overall moderate CO_2 source activity, related to a shorter period of photosynthetic uptake and metabolic costs of N_2 fixation. Perennial legumes (e.g., alfalfa, *Medicago sativa* L.) were strong sinks for atmospheric CO_2, with an average net ecosystem production of 980 (range 550–1200) (kg ha^1). The variable increase in crop yield, and thus in NPP with BSA treatments, was similar to wheat grown in a temperate loam soil (Vaccari et al., 2011). The insignificant increase in soil respiration and a potential increase in crop yield with BSA suggest that biochar has the potential for enhancing ecosystem CO_2 uptake and crop productivity in regions with low fertility soil and poor agricultural production. The results of our study showed that biochar, in combination with chemical fertilizer, have the potential to increase BNF in legumes, crop productivity and reduce CO_2 emissions in legume-cereal crop rotations on arid soils.

5. Conclusion

The present study improves our understanding of net ecosystem CO_2 exchange and soil carbon status affected by biochar applications in legume-cereal cropping systems of a rainfed area. Biochar applications increased SOC, TN and crop yield, and decreased soil bulk density. Increased CO_2 emissions were observed from mash bean plots that were amended with biochar in the presence of NPK. However, biochar applications significantly improved N_2-fixation and increased nodule number in both growing seasons. This study suggests that biochar amendments could improve the quality of calcareous soils that are naturally low in organic carbon, and in turn, enhance mash bean and wheat production. Our results show that the use of sugarcane-bagasse biochar as a soil amendment could be an effective strategy for achieving high grain yield of both crops while lowering the global warming potential over a longer period of time.

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